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EVALUATION OF ANTHELMINTIC ACTIVITY OF CUCUMIS PUBESECENS METHANOLIC EXTRACT

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Abstract

Helminthiasis, also known as worm infection, is any macro parasitic disease of humans and other animals in which a part of the body is infected with parasitic worms, known as helminths. *Cucumis pubescens*, also known as kachri, has various therapeutic effects in gastrointestinal pain, used as antimicrobial, antioxidant, anti-inflammatory and supports immune function. Phytochemical screening of methanolic extract of *Cucumis pubescens* fruits was carried out using different reagents, which revealed the presence of alkaloids, flavonoids, terpenoids, steroids, tannins, glycosides. Methanolic extract of *Cucumis pubescens* fruits (MECP) was investigated for their anthelmintic activity against *Pheretima posthuma*. Various concentrations (1.25-10 mg/ml) of each extract were tested in the bioassay, which involved determination of time of paralysis and time of death of the worms.

Keywords: Helminthiasis, *Cucumis pubescens*, MECP, anthelmintic, *Pheretima posthuma*.

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Introduction

Helminthiasis, also known as worm infection, is any macro parasitic disease of humans and other animals in which a part of the body is infected with parasitic worms, known as helminthes [1,2]. Helminth infections are among the commonest infections in man, affecting a large proportion of the world's population. In developing countries they pose a major threat to public health and contribute to the prevalence of malnutrition, anemia, eosinophilia, and pneumonia [2-4]. Anthelmintics are drugs that either kill or expel infesting helminths and the gastrointestinal tract is the abode of many helminths, although some also live in tissues, or their larvae migrate into tissues [5]. They harm the host by depriving him of food, causing blood loss, injury to organs, intestinal or lymphatic obstruction and by secreting toxins. Helminthiasis is rarely fatal, but is a major cause of morbidity [6,7].

Cucumis pubescens, also known as kachri, is a wild cucumber plant with hairy stems, yellow flowers, and fruit that can be yellow, brown, or striped green. It is a member of the Cucurbitaceae family. The chemical components of *Cucumis pubescens* include flavonoids, alkaloids,

glycosides, phenolics, tannins, terpenoids, carotenoids, steroids, Saponins and resins. It has various therapeutic effects in gastrointestinal pain, used as antimicrobial, antioxidant, anti-inflammatory and supports immune function [8]. The primary objective of the present study as to evaluate Anthelmintic activity of methanolic extract of *Cucumis pubescens* fruit by in-vitro method.

Materials & Methods

Collection of the plant

The plant *Cucumis pubescens* was collected from the cultivation field in Chittoor in the month of September 2024. The plant had been authenticated by botanist, Dr. N. Sivaraj.

Extraction of the plant material

The fruits of *Cucumis pubescens* were shade dried and coarsely powdered. The powder was extracted with methanol by continuous hot extraction using soxhlet apparatus for 72 h [9]. The extract obtained was concentrated in a rotary evaporator at a temperature less than 45° and preserved in desiccators for further use [10,11].

Phytochemical screening of the extract

Phytochemical screening was carried out using differential reactions (staining and precipitation) of the main groups of chemical compounds contained in the plants according to the classical methods. The samples were analyzed for

the presence of different classes of secondary metabolites including flavonoids, alkaloids, tannins, glycosides, saponins, triterpenes, and phenols [12, 13].

Invitro anthelmintic activity of methanolic extract of *Cucumis pubescens* fruits (MECP)

Collection of Earthworms

Earthworms (*Pheretima posthuma*) were collected from the swampy water near kalavagunta dam, Muthirevula, Chittoor, Andhra Pradesh, India. The average size of worms was 5-8cm. The anthelmintic activity was carried out as per the method described. The assay was performed in vitro using adult earthworms owing to their anatomical and physiological resemblance with the intestinal round worms, parasites of human beings for preliminary evaluation of anthelmintic activity. Concentration of standard drug (Albendazole) and methanolic extract of *Cucumis pubescens* fruits were prepared [14, 15].

Evaluation of Anthelmintic activity using Earthworms

Earthworms, each of average length of 6cm, were placed in petridishes containing 10mL of various drug concentrations, 0.25mg/ml of Albendazole and 1.25 mg/mL, 2.5 mg/mL, 5mg/mL, 10mg/mL, of methanolic extract of *Cucumis pubescens* fruits. Albendazole solution was used as reference standard drug and phosphate buffer with pH 7.4 as control. The worms were observed for the motility after incubating at 37°C. This was done after pouring the Petri dishes content in the wash basin and allowing the worms to move freely. By tapping the end of each worm with the index finger and applying a bit of pressure, the worms that were alive showed motility and those dead were non motile [16].

The motile worms were returned to the respective petridishes containing drug solutions, and the incubation process was carried out again. In the control, the worms were available for at least twelve days, which is similar to the findings reported earlier. The time taken for paralysis, motility activity of any sort, and death time of worms were observed and recorded after ascertaining that the worms did not move neither when shaken vigorously nor when dipped in warm water [17, 18].

Statistical analysis: All experiments were repeated at least three times and the results expressed as mean \pm S.E.M. The statistical analysis of data was done using one-way ANOVA (Analysis of Variance) with level of statistical significance taken as $p < 0.05$.

Results & Discussion

Phytochemical screening of the extract

Preliminary phytochemical screening of methanolic extract of *Cucumis pubescens* fruits revealed the presence of alkaloids, flavonoids, terpenoids, steroids, tannins, glycosides (Table 1).

Table 1. Phytochemical screening of MECP

S.No	Phytoconstituent	MECP
1	Alkaloids	+
2	Flavonoids	+
3	Proteins	-
4	Terpenoids	+
5	Steroids	+
6	Tannins	+
7	Glycosides	+
8	Phenol	-
9	Saponins	-

+ indicates presence, - indicates absence

Invitro anthelmintic activity of methanolic extract of *Cucumis pubescens* fruits (MECP)

Methanolic extract of *Cucumis pubescens* fruits (MECP) was investigated for their anthelmintic activity against *Pheretima posthuma*. Various concentrations (1.25-10 mg/ml) of each extract were tested in the bioassay, which involved determination of time of paralysis and time of death of the worms. Observations were made for the time taken to paralyze and death of individual worms. Time for paralysis was noted when no movement could be observed except when the worms were shaken vigorously. Death was concluded when the worms lost their motility followed with fading away of their body colors (Table 2 & Figure 1).

In vitro anthelmintic activity was performed and the paralysis time and lethal time were recorded. Statistical evaluation of the data was performed by one-way ANOVA. The results were expressed as mean \pm SD using Graph Pad.

Table 2. In vitro anthelmintic activity of MECP &

Albendazole

S.No	Groups	Paralysis time (min)	Death time (min)
1	PBS	-	-
2	Albendazole 0.25mg/ ml	34 \pm 0.02**	67 \pm 0.01**
3	MECP 1.25mg/ml	97 \pm 0.04	198 \pm 0.02
4	MECP 2.5mg/ml	71 \pm 0.02	141 \pm 0.01
5	MECP 5mg/ml	56 \pm 0.03*	96 \pm 0.02*
6	MECP 10mg/ml	20 \pm 0.01**	32 \pm 0.03**

Values are expressed as mean \pm SEM. If $P < 0.05$ is considered as statistically significant

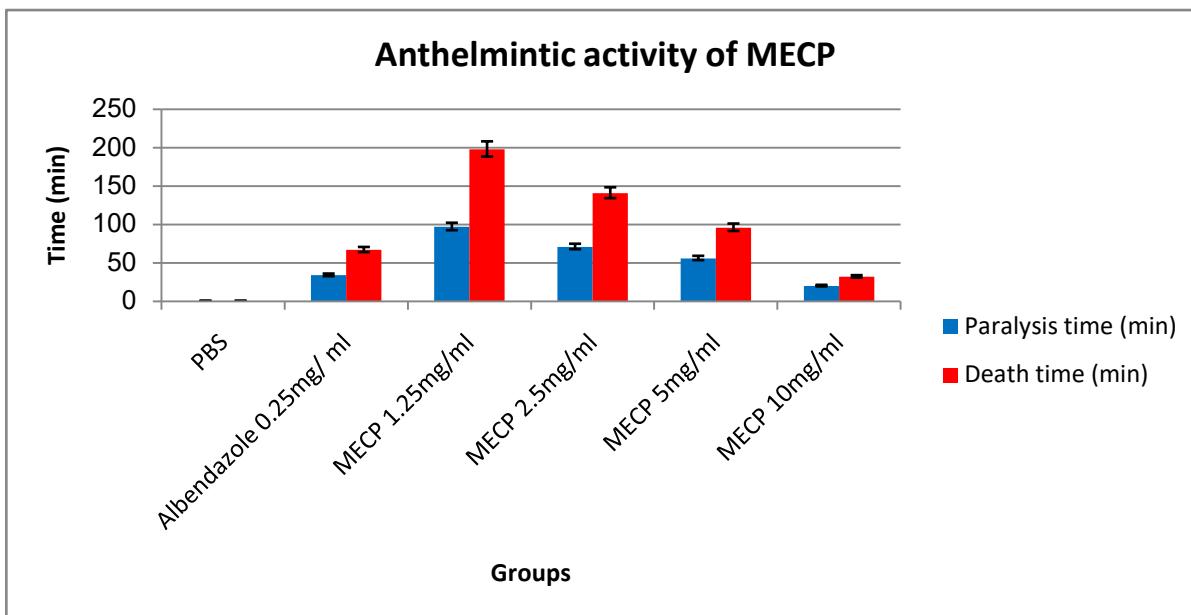


Figure 1. Anthelmintic activity of MECP

Conclusion

As per the results shown in Table 1, the predominant effect of Methanolic extract of *Cucumis pubescens* fruits (MECP) on the worm was to cause a flaccid paralysis that resulted in expulsion of the worm by peristalsis. MECP by increasing chloride ion conductance of worm muscle membrane produces hyperpolarization and reduced excitability that leads to muscle relaxation and flaccid paralysis. The extract showed significant anthelmintic activity on selected worms. MECP found to be significantly active as compared to standard. Further studies are necessary to identify the chemical moieties responsible for anthelmintic activity and also to evaluate mechanism of action at cellular and molecular level.

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Conflict of Interest

The authors declare no conflict of interest.

Informed Consent

NA

Ethical Statement

NA

Author Contribution

Concept: G. Naveen Kumar, design: G. Naveen Kumar, data collection: Chittibabu Lakshmi Priya, Aniket Kumar, Bugude Vamsi Kumar, P Raghu, Sachin Kumar, analysis: D. Jothieswari, writing: Chittibabu Lakshmi Priya, Aniket Kumar, Bugude Vamsi Kumar, P Raghu, Sachin Kumar.

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